PATENT COOPERATION TREAT

From the
INTERNATIONAL PRELIMINARY EXAMINING AUTHORITY

To:
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PCT

NOTIFICATION OF TRANSMITTAL OF INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY
(Chapter II of the Patent Cooperation Treaty)

(PCT Rule 71.1)

Date of mailing *(day/month/year)*	22 February 2006 (22-02-2006)
Applicant's or agent's file reference 82008WO0	**IMPORTANT NOTIFICATION**

International application No.	International filing date *(day/month/year)*	Priority date *(day/month/year)*
PCT/CA2004/001870	25 October 2004 (25-10-2004)	24 October 2003 (24-10-2003)

Applicant
UNIVERSITY OF SASKATCHEWAN ET AL

1. The applicant is hereby notified that this International Preliminary Examining Authority transmits herewith the international preliminary report on patentability and its annexes, if any, established on the international application.

2. A copy of the report and its annexes, if any, is being transmitted to the International Bureau for communication to all the elected Offices.

3. Where required by any of the elected Offices, the International Bureau will prepare an English translation of the report (but not of any annexes) and will transmit such translation to those Offices.

4. **REMINDER**

The applicant must enter the national phase before each elected Office by performing certain acts (filing translations and paying national fees) within 30 months from the priority date (or later in some Offices) (Article 39(1)) (see also the reminder sent by the International Bureau with Form PCT/IB/301).

Where a translation of the international application must be furnished to an elected Office, that translation must contain a translation of any annexes to the international preliminary report on patentability. It is the applicant's responsibility to prepare and furnish such translation directly to each elected Office concerned.

For further details on the applicable time limits and requirements of the elected Offices, see Volume II of the *PCT Applicant's Guide.*

The applicant's attention is drawn to Article 33(5), which provides that the criteria of novelty, inventive step and industrial applicability described in Article 33(2) to (4) merely serve the purposes of international preliminary examination and that "any Contracting State may apply additional or different criteria for the purposes of deciding whether, in that State, the claimed invention is patentable or not" (see also Article 27(5)). Such additional criteria may relate, for example, to exemptions from patentability, requirements for enabling disclosure, clarity and support for the claims.

Name and mailing address of the IPEA/CA	Authorized officer
Canadian Intellectual Property Office Place du Portage I, C114 - 1st Floor, Box PCT 50 Victoria Street Gatineau, Quebec K1A 0C9 Facsimile No.: 001(819)953-2476	Wendy McQuaig (819) 994-3711

PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY
(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 82008WO0	FOR FURTHER ACTION	See Form PCT/IPEA/416
International application No. PCT/CA2004/001870	International filing date *(day/month/year)* 25 October 2004 (25-10-2004)	Priority date *(day/month/year)* 24 October 2003 (24-10-2003)

International Patent Classification (IPC) or national classification and IPC
IPC: A61K 48/00 ***(2006.01)*** , A61P 17/00 ***(2006.01)*** , A61K 47/44 ***(2006.01)*** , A61K 38/21 ***(2006.01)*** , A61K 31/7088 ***(2006.01)*** , A61K 9/06 ***(2006.01)***

Applicant
UNIVERSITY OF SASKATCHEWAN ET AL

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 7 sheets, including this cover sheet.

3. This report is also accompanied by ANNEXES, comprising:

   a. [X] *(sent to the applicant and to the International Bureau)* a total of 2 sheets, as follows:

      [X] sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).

      [ ] sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.

   b. [ ] *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s))
      , containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

   [X] Box No. I Basis of the report
   [ ] Box No. II Priority
   [ ] Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   [ ] Box No. IV Lack of unity of invention
   [X] Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   [ ] Box No. VI Certain documents cited
   [ ] Box No. VII Certain defects in the international application
   [X] Box No. VIII Certain observations on the international application

Date of submission of the demand	Date of completion of this report
10 August 2005 (10-08-2005)	22 February 2006 (22-02-2006)
Name and mailing address of the IPEA/CA Canadian Intellectual Property Office Place du Portage I, C114 - 1st Floor, Box PCT 50 Victoria Street Gatineau, Quebec K1A 0C9 Facsimile No.: 001(819)953-2476	Authorized officer Debora Fujimoto (819) 997-1855

Box No. I Basis of the report

1. With regard to the **language**, this report is based on:

[X] the international application in the language in which it was filed

[ ] a translation of the international application into , which is the language of a translation furnished for the purposes of:

[ ] international search (Rules 12.3(a) and 23.1(b))

[ ] publication of the international application (Rule 12.4(a))

[ ] international preliminary examination (Rules 55.2(a) and/or 55.3(a))

2. With regard to the **elements** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report):*

[ ] the international application as originally filed/furnished

[X] the description:

[X] pages 1-34 as originally filed/furnished

[ ] pages* received by this Authority on

[ ] pages* received by this Authority on

[X] the claims:

[ ] pages as originally filed/furnished

[ ] pages* as amended (together with any statement) under Article 19

[X] pages* 35-36 received by this Authority on 30.08.2005

[ ] pages* received by this Authority on

[X] the drawings:

[X] pages 1/7-7/7 as originally filed/furnished

[ ] pages* received by this Authority on

[ ] pages* received by this Authority on

[X] a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing.

3. [X] The amendments have resulted in the cancellation of:

[ ] the description, pages

[X] the claims, Nos. 20-100

[ ] the drawings, sheets/figs

[ ] the sequence listing *(specify):*

[ ] any table(s) related to sequence listing *(specify):*

4. [ ] This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

[ ] the description, pages

[ ] the claims, Nos.

[ ] the drawings, sheets/figs

[ ] the sequence listing *(specify):*

[ ] any table(s) related to sequence listing *(specify):*

* *If item 4 applies, some or all of those sheets may be marked "superseded."*

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims	1-19	YES
	Claims		NO
Inventive step (IS)	Claims		YES
	Claims	1-19	NO
Industrial applicability (IA)	Claims	1-19 (partially)	YES
	Claims	1-19 (partially)	NO

2. Citations and explanations (Rule 70.7)

D1 to D5 are identical to those previously cited in the International Search Report and the Written Opinion.

D1 McGREGOR C ET AL. Rational approaches to the design of cationic gemini surfactants for gene delivery. J AM CHEM SOC 4 Jul 2001 Vol 123, No 26, pages 6215-6220
D2 RONSIN G ET AL. Novel spermine-based cationic gemini surfactants for gene delivery. CHEM COMMUN (CAMB) 7 Nov 2001 Vol 7, No 21, pages 2234-2235
D3 WO 9929712 A1 (SMITHKLINEBEECHAM PLC) 17 Jun 1999
D4 VOGEL JC. Nonviral gene therapy. HUMAN GENE THERAPY 1 Nov 2000 Vol 11, pages 2253-2259
D5 SAPADIN AN & FLEISCHMAJER R. Treatment of scleroderma. ARCH DERMATOL Jan 2002 Vol 138, No 1, pages 99-105

The problem solved by the instant application is the provision of a topical delivery system, comprising a biologically active agent and a cationic gemini surfactant, and use thereof for application to the skin or mucosal membrane to generate a localized or systemic therapeutic effect.

D1 compares the use of cationic gemini surfactants to the cationic lipid composition, LipofectAMINE PLUS™, for transfection of a luciferase reporter gene into CHO-DG44 (Chinese Hamster Ovary), C2C12 mouse muscle, and human neuronal cells. Increasing the length of the hydrocarbon tail of the surfactant from C12 to C18 leads to a substantial increase in gene expression (Fig. 1b). Figure 2 and page 6217 disclose that when compared to the use of the cationic gemini surfactant alone, the use of the combination of the gemini surfactant GS11 and one of the supplements, polylysine or LipofectAMINE PLUS™, significantly increases gene expression efficiency in different cell types. The preparation of a delivery system comprising the gemini surfactant GS11 and the supplement, dioleoyl phosphatidylethanolamine (DOPE), is specifically disclosed (page 6217, second column through page 6218, first column).

D2 discloses that the use of two of four cationic gemini surfactants tested for their ability to transfect cell lines (CHO, neuronal, mouse muscle and mouse tumour) with a luciferase reporter gene results in better transfection in all the cell lines when compared to the use of LipofectAMINE™ 2000, a cationic lipid composition. The gene expression frequency resulting from the addition of polylysine to the delivery system comprising a cationic gemini surfactant depends on the selected cationic gemini surfactant. The results suggest that the selection of a gemini surfactant should be tailored for use with the targeted cell type.

D3 demonstrates the use of two of five cationic gemini compounds alone results in a higher transfection efficiency of a luciferase recombinant plasmid into HEK 293 cells (Example 17) than the use of LipofectAMINE™ or LipoTAXI™, cationic lipid compositions. In contrast, use of an anionic gemini compound resulted in a transfection frequency comparable to the "no DNA" control. Similar results were obtained in CHO-K1 (Chinese hamster ovary) cells (Example 18), demonstrating good transfection efficiency when cationic gemini compounds are used and negligible transfection efficiency when an anionic gemini compound is used. D3 additionally discloses that the neutral carrier DOPE, peptides, basic amino acids, and complexing agents such as the PLUS™ reagents (page 5) comprise the group of supplements that are known to increase transfection efficiency. D3 specifically discloses that the use of a cationic gemini compound in combination with the supplement selected from (i) DOPE, (ii) LipofectAMINE PLUS™ and (iii) DOPE and LipofectAMINE PLUS™ results in a higher transfection frequency of a luciferase recombinant plasmid into HEK293 (hamster embryonic kidney) cells than the use of a cationic gemini compound alone (Example 19).

(Continued in Supplemental Box)

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

Claim Defects:
Claims 1and 2 do not comply with Article 6 of the PCT. The only type of gemini surfactant used successfully in the present application is cationic. Thus, there is no support in the description for the topical delivery system and use thereof of gemini surfactants selected from anionic, neutral, amphoteric, or mixtures thereof referred to in claim 2.

Claim 1 does not comply with Article 6 of the PCT. The phrase "biologically active agent" is vague and indefinite, and thus, claim 1 lacks clarity.

Supplemental Box relating to Sequence Listing

Continuation of Box No.1, item 2:

1. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, this report was established on the basis of:

   a. type of material

      [X] a sequence listing

      [ ] table(s) related to the sequence listing

   b. format of material

      [X] on paper

      [X] in electronic form

   c. time of filing/furnishing

      [X] contained in the international application as filed

      [X] filed together with the international application in electronic form

      [ ] furnished subsequently to this Authority for the purposes of search and/or examination

      [ ] received by this Authority as an amendment* on

2. [X] In addition, in the case that move than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

* *If item 4 in Box No. 1 applies, the listing and/or table(s) related thereto, which form part of the basis of the report, may be marked "superseded".*

Supplemental Box

In case **the space in any of the preceding boxes is not sufficient.**

Continuation of: **Box No. V**

D4 discloses the topical application of liposome-coated DNA for therapy of skin diseases, wherein the DNA codes for growth factors, cytokines, and structural genes. D4 additionally discloses that the delivery of liposome-coated DNA to induce local expression of cytokines (IL-12, IL-18, IL-1-β, IL-6, and IFN-γ), growth factors, or foreign major histocompatibility complex proteins is used for antitumor therapy. Further disclosed are other methods for delivery of DNA using biolistic methods, direct injection of immunostimulatory CpG-containing oligodeoxynucleotides (CpG/ISS-ODN), and electroporation of antisense ODN for anti-tumor therapy and of chimeric RNA/DNA-ODN for therapy of skin diseases, wound healing and hair follicle manipulation. Therefore, D4 discloses the use of a variety of biologically active agents, including DNA encoding IFN-γ, to produce a therapeutic effect.

D5 discloses that interferon-γ has been shown to be an antifibrotic agent with mild beneficial effects on skin sclerosis and disease-associated symptoms during clinical trials.

Novelty:
D1 compares the transfection frequency of plasmid DNA in different cell types that results from the use of the delivery system comprising a cationic gemini surfactant and the supplement, DOPE, to that of the use of the delivery system comprising a cationic lipid composition. However, D1 does not specifically disclose the system comprising a gemini surfactant and a DNA encoding interferon-γ for topical delivery to generate a therapeutic effect.

D2 discloses that cationic gemini surfactants are used to transfect cell lines with plasmid DNA. D2 does not disclose the topical delivery system comprising the DNA encoding interferon-γ used in combination with a gemini surfactant to generate a therapeutic effect.

D3 compares the transfection efficiencies of a plasmid DNA in different animal cell lines using a cationic gemini surfactant alone, or in combination with supplements such as DOPE and LipofectAMINE PLUS™. D3 does not disclose the specific delivery system comprising a biologically active agent, DNA encoding interferon-γ, and a gemini surfactant for topical application to skin or a mucosal membrane to generate a therapeutic effect.

D4 discloses that DNA encoding a product is deliverable by a number of therapeutic methods. D4 specifically discloses the topical application of DNA encoding a cytokine in a liposomal composition for anti-tumor therapy. D4 does not disclose the use of a liposomal delivery system comprising a gemini surfactant and DNA encoding a cytokine for topical application to skin or a mucosal membrane to generate a therapeutic effect.

D5 discloses the use of interferon-γ for the treatment of skin disorders and diseases arising from an interferon-γ deficiency. D5 does not specifically disclose the use of a topical delivery system comprising a gemini surfactant in combination with DNA encoding interferon-γ to generate a therapeutic effect.

In view of any one of D1-D5 taken independently, the subject matter of claims 1-19 appears to be novel and complies with Article 33(2) of the PCT.

Inventive Step:
Applicant's arguments in his letter of 30 August 2005 have been taken into consideration. In the response to the written opinion dated 30 August 2005, applicant argues that "D4 nowhere mentions any specific formulations for topical skin delivery of DNA, and certainly makes no mention of gemini surfactants as being capable of topical delivery". Applicant refers to the passage in D4 that states the "stratum corneum, which is the outer protective layer of the epidermis, is hydrophobic and presents a formidable barrier to large negatively charged molecules such as DNA, even when complexed to liposomes" as proof that one would not be motivated to use a gemini surfactant in a topical delivery system. D4 demonstrates on page 2254 that DNA-liposomal formulations are well known in the art, and Tables 1 and 2 of D4 disclose the use of said DNA-liposomal formulations for topical delivery and expression of cytokines. Additionally, referring to the DNA-liposome topical method for gene delivery, D4 further states that although "[O]nly epidermal cells would be targeted with this delivery method and expression would be relatively low and transient ... this delivery method may be effective for immunization and treatment of skin disorders localized to the hair follicle" (page 2254, second col.). Therefore, D4 discloses the feasability of using topically applied DNA-liposomal compositions to treat skin disorders.

(Continued in Supplemental Box)

Supplemental Box

In case **the space in any of the preceding boxes is not sufficient.**

Continuation of: **Box No. V**

The objections for lack of inventive step in the present application are maintained in view of D1 to D5, as discussed below.

It is obvious in view of D1, which compares delivery systems comprising known cationic liposomal compositions such as LipofectAMINE PLUS™, LipofectAMINE™ 2000, or LipoTAXI™ to delivery systems comprising cationic gemini surfactants, that the two types of delivery systems are designed for the same purpose, i.e., gene delivery into different cell types. Further, in view of D4, it is obvious to one skilled in the art to use supplements, such as DOPE, that are known to enhance transfection efficiency when used in combination with cationic liposomal compositions to enhance transfection efficiency when used in combination with the cationic gemini surfactants of the present application. Therefore, D1 discloses a delivery system comprising a cationic gemini surfactant and a biologically active agent but not the therapeutic use of said delivery system by topical application to the skin and mucosal membranes. D4 discloses different methods for gene delivery, including the topical application of liposome-coated DNA coding for a cytokine to the skin to generate a therapeutic effect. D5 discloses clinical trials in which the cytokine, interferon-γ, is used to treat scleroderma. One of skill in the art would be motivated by the disclosures of D4 and D5 to use a topical delivery system comprising interferon-γ to treat a skin disorder. Therefore, it is obvious to one of skill in the art to substitute the cationic lipid composition with a cationic gemini surfactant disclosed in D1 in a topical delivery system to deliver DNA encoding the cytokine IFN-γ for the therapeutic treatment of a skin disorder in view of D4 and D5, taken together. The subject matter of claims 1-19 appears to lack an inventive step in view of D1, D4, and D5 taken together, and does not comply with Article 33(3) of the PCT.

D2 discloses the use of a delivery system comprising a cationic gemini surfactant and a supplement to deliver DNA to a variety of cell line types, and that the effects and use of said cationic gemini surfactant are comparable to known cationic liposomal compositions such as LipofectAMINE™ 2000. D4 discloses different methods for gene delivery, including the topical application of liposome-coated DNA coding for a cytokine to the skin to generate a therapeutic effect. D5 discloses clinical trials in which the cytokine, interferon-γ, is used to treat scleroderma. In combination, D4 and D5 suggest that a delivery system comprising interferon-γ has a known use for topical gene delivery to treat skin disorders. Therefore, it is obvious to one of skill in the art to substitute a cationic lipid composition with a cationic gemini surfactant in a topical delivery system to deliver DNA encoding the cytokine IFN-γ to the skin or mucosal membrane to generate a therapeutic effect in view of D2, D4, and D5, taken together. The subject matter of claims 1-19 appears to lack an inventive step in view of D2, D4, and D5 taken together, and does not comply with Article 33(3) of the PCT.

D3 discloses a delivery system comprising a cationic gemini surfactant and a biologically active agent. As D3 also discloses that the effects and use of the cationic gemini surfactant for transfection of cells are comparable to known cationic compositions such as LipofectAMINE PLUS™, LipofectAMINE™ 2000, and LipoTAXI™, it is obvious to one skilled in the art to substitute the known cationic compositions with the cationic gemini surfactant disclosed in D3. Further, D3 discloses that inclusion of a supplement such as DOPE in a delivery system for the transfection of animal cells is known to increase transfection efficiency. D4 discloses different methods for gene delivery, including the topical application of liposome-coated DNA coding for a cytokine to the skin to generate a therapeutic effect. D5 discloses clinical trials in which the cytokine interferon-γ is used to treat scleroderma. In combination, D4 and D5 suggest that a delivery system comprising interferon-γ has a known use for topical gene delivery to treat skin disorders. Therefore, it is obvious to one skilled in the art to use a cationic gemini surfactant in a topical delivery system to deliver DNA encoding the cytokine IFN-γ to the skin or mucosal membranes to generate a therapeutic effect in view of D3-D5, taken together. The subject matter of claims 1-19 appears to lack an inventive step in view of D3-D5 taken together, and does not comply with Article 33(3) of the PCT.

Industrial Applicability:
D3 discloses that the use of an anionic gemini surfactant to transfect a plasmid expressing luciferase into cell lines results in a transfection efficiency comparable to the "no DNA added" control, and only two of five cationic gemini surfactants used to transfect the identical plasmid results in higher transfection efficiency. Therefore, it appears that not all gemini surfactants function equally to facilitate the delivery of plasmid DNA. In view of D3, claims 1-19 appear to define subject matter that has partial industrial applicability when referring to cationic gemini surfactants, under Article 33(4) of the PCT.

WE CLAIM:

1. A topical delivery system, comprising:

a gemini surfactant in admixture with a biologically active agent, wherein the delivery system, when in contact with skin or a mucosal membrane, provides a localized or systemic therapeutic effect.

2. The delivery system according to claim 1, wherein the gemini surfactant is selected from an anionic gemini surfactant, a gemini cationic surfactant, a neutral gemini surfactant, an amphoteric gemini surfactant, or mixtures thereof.

3. The delivery system according to claim 1 or claim 2, wherein the gemini cationic surfactant has a hydrophobic tail comprising a C_3-C_{30} alkyl group.

4. The delivery system according to any one of claims 1-3, wherein the biologically active agent is a plasmid DNA.

5. The delivery system according to claim 4, wherein the plasmid DNA comprises a gene encoding for interferon-γ.

6. The delivery system according to any one of claims 1-5, wherein the delivery system includes one or more pharmaceutically-acceptable vehicles.

7. The delivery system according to any one of claims 1-5, wherein the delivery system includes one or more supplements suitable for application for skin or mucosa.

8. The delivery system according to claim 6 or claim 7, wherein the delivery system is in the form of a cream, lotion, paste, ointment, foam, gel, lipid formulation, emulsion, solution, or suspension.

9. The delivery system according to claim 8, further comprising one or more supplements selected from a neutral carrier or a permeation enhancer.

10. The delivery system according to claim 9, wherein the neutral carrier is selected from 1,2-dioleyl-sn-glycero-phosphatidylethanolamine (DOPE) or cholesterol.

11. The delivery system according to claim 8, further comprising a compound selected from diethylene glycol monoethyl ether, polyglyceryl 3-diisostearate, PEG-8 caprylic and capric glycerides, and octyldodecyl myristate.

12. The delivery system according to any one of claims 1-5, in the form of a liquid formulated as an aqueous solution, non-aqueous solution, aerosol, mist, spray, drops, or instillation.

13. Use of the topical delivery system according to any one of claims 1-12 for treatment of a skin disorder.

14. The use according to claim 13, for treatment of scleroderma, atopic dermatitis, or psoriasis.

15. The use according to claim 13, wherein the skin disorder is of genetic origin.

16. Use of the topical delivery system according to any one of claims 1-12 for treatment of a metabolic disease selected from the group consisting of gyrate atrophy, maternal hyperphenylalaninemia, familial hypercholesterolemia, and phenylketonuria.

17. Use of a gemini surfactant in the manufacture of a topical delivery system according to any one of claims 1-12.

18. The use according to claim 17 for manufacture of a medicament comprising as the biologically active agent a plasmid DNA comprising the gene encoding for interferon-γ, said medicament suitable for treatment of scleroderma, atopic dermatitis, or psoriasis.

19. Use of the topical delivery system according to any one of claims 1-12 for treatment of a systemic protein deficiency.